# Studies on mass culture and aggregation pheromones in the exotic powderpost beetle, Lyctus africanus Lesne (Coleoptera: Lyctinae)( Abstract_要旨 )

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（ 続紙 1 ）

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| 論文題目 | Studies on mass culture and aggregation pheromones in the exotic powderpost beetle, *Lyctus africanus* Lesne (Coleoptera: Lyctinae)<br>（外来木材害虫アフリカヒラタキクイムシの大量飼育および集合フェロモンに関する研究） | | |

（論文内容の要旨）

Several species of the wood pest Lyctinae subfamily, belonging to the Bostrichidae family, have been introduced in countries such as Australia, New Zealand, USA/Canada, Europe, countries around the Mediterranean and Japan. Though the Lyctinae are distributed in all the faunal regions, the beetles are supposed to be an endemic species of oriental and Ethiopian regions. In Japan, some Lyctinae, *Lyctus africanus, L. brunneus*, *Minthea rugicollis, L. linearis,* and *Lyctoxylon dentatum* had established in some prefectures and being registered as alien species. Furthermore, *L. africanus* was discovered frequently in this country rather than the other species. Nowadays, the cases of *L. africanus* are increasing in Japan.

Recently, *L. africanus* has become an important pest owing to its migration into new countries, and it has been considered to be one of the major pests threatening timber and wood products, including plywood, dried roots, seeds and tubers. This beetle attacks sawn hardwood timber products and also damage many manufactured wood products. It may also infest dried roots and tubers.

Lyctine damage is mostly identified belatedly by reason of poor knowledge and skill to locate and monitor the infestation. Some monitoring techniques have been developed to regularly inspect insect populations, such as an insect trap. Unfortunately, there is still no device for detecting and monitoring Lyctine beetles in the field due to the lack of sufficient data on their ecological features. Thus, strategies for monitoring and controlling this beetle by examining the Lyctine ecology are urgently needed. To accomplish the study, establishment of the mass culture of *L. africanus* was also elaborated. This study aims at the establishment of the mass culture and monitoring system for *L. africanus*.

To establish the mass culture of *L. africanus*, the significance of the food or artificial diet quality toward fecundity and also population of the *Lyctus* were discussed in Chapter 1 and Chapter 2. The studies were emphasized on finding the standard diet for rearing *L. africanus* in laboratory. By understanding the favorable diet composition in *L. africanus*, it must become possible to supply more *Lyctus* adult in laboratory for further study on monitoring of *L. africanus* infestation.

In experiments, the usefulness of some artificial diets with different fillers was

evaluated to improve the growth of *L. africanus*. *L. africanus* were raised in three types of diets (wood– [Diet 1], cellulose powder– [Diet 2], and alpha-cellulose– [Diet 3] based diet). A group of five females and males *L. africanus* was liberated onto the diet and allowed to complete the life cycle. The new generations were harvested and then subjected to several experiment, such as oviposition test, total population measurement and also determination of sex ratio and body weight of the beetle.

In Chapter 1, fecundity of *L. africanus* by concerning oviposition ability was discussed. The oviposition test was conducted on a folded filter paper. The numbers of laid eggs and survived adults after oviposition test were observed. The results revealed that combination of starch and sugar acted as oviposition stimulant for *Lyctus* adults emerged from Diet 1, Diet 2, and Diet 3. The adults emerging from cellulose-based diets (Diet 2 and Diet 3) were likely to oviposit more eggs than what Diet 1-emerged adult did, which indicated that cellulose might support the oviposition ability of *L. africanus*. However, all adults emerging from three diets survived in the same values after laying eggs on both oviposition sites.

Furthermore, the newly emerged beetles were harvested and observed the total population of larvae, adults, as well as sex ratio and body weight measurements, as described in Chapter 2. The results indicated that total larvae, sex ratio, and body weight of *L. africanus* were similar among the three diets. However, the number of total adults was significantly lower in the Diet 3 than the other diets. It was suggested that the amount of vital nutrient (starch) in the diet is not the only important factor to be considered when selecting a diet for *L. africanus*. The filler should also enhance oviposition potential and larval development. In this study, Diet 1 and Diet 2 could be used alternately for rearing *L. africanus* in laboratory instead of Diet 3.

Chapter 3 discussed the aggregation behavior of *L. africanus* in order to develop a monitoring technique for the beetle. The behavior was investigated using a chemical approach through comprehensive screening of the potential compound produced by *L. africanus.*

The whole body extractions using hexane solvent on newly emerged beetles were performed. The aggregation activity of crude extract of *L. africanus* beetles was conducted by dual-choice bioassay. Then, it was followed by chemical analysis, isolation, identification and syntheses of the chemical compounds. The results revealed three esters as aggregation pheromones produced by male *L. africanus* beetle. The esters were recognized as a major compound (3-pentyl dodecanoate) and two minor compounds (2-propyl dodecanoate and 3-pentyl tetradecanoate) based on its quantitative amount in the crude extract.

In order to justify the significant role of the synthetic esters to *L. africanus* the

aggregation activity of either single or blended esters was discussed in Chapter 4. The bioassays were conducted in laboratory by dual-choice test against *L. africanus* beetles. The results indicated that the single compound was not sufficient to induce the aggregation behavior of *L. africanus*. Furthermore, the natural blended compound increased the aggregation responses of *L. africanus*. A synergistic effect was found among the three synthetic esters.

A semi-field test was carried out in order to initiate a pheromone-based monitoring program in the future, as discussed in Chapter 5. The optimum dose of three-ester blend was determined using a dual-choice test. The blend of three synthetic esters was prepared to determine the minimum dosage that elicited the maximum level of response from both female and male *L. africanus* beetles. Consequently, the optimum dose inducing greatest aggregation response on *L. africanus* was applied for a wind-tunnel bioassay.

Results of dual-choice bioassays indicated that 50 Beetle Equivalent (BE) induced the greatest response of aggregation behaviour of *L. africanus*. In addition, the wind tunnel bioassays revealed that both female and male beetles showed arousal response toward the natural blend of esters, however the male were less responsive than the female *L. africanus* beetles. The aggregation pheromone might efficiently perform on female rather than male *L. africanus* beetles.

These results indicated the potential ability of ester compounds as aggregation pheromones of *L. africanus* to be developed as an attractant in insect monitoring. Additional studies are still necessary to strengthen the performance of synthetic esters for establishing the monitoring system of *L. africanus*.

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（続紙 ２ ）

（論文審査の結果の要旨）

海外からの木材輸入が丸太から製品へと大きく変化し続ける中、乾燥木材害虫の侵入・拡大リスクが高まりつつある。特にヒラタキクイムシ類とナガシンクイ類は、世界的にも経済的な重要性が高い害虫である。これらは熱帯に多くの種類が分布し、辺材を含む熱帯産広葉樹製品とともに持ち込まれる可能性がある。事実、西日本では、従来最重要種であると考えられていたヒラタキクイムシからアフリカを原産地とするアフリカヒラタキクイムシへと害虫種の変化が生じつつあることが知られている。本論文は、外来木材害虫アフリカヒラタキクイムシの大量飼育法の確立と、化学生態学的手法を利用した新たな防除技術の開発を目指したものであり、評価すべき点として以下の３点を挙げることができる。

１．アフリカヒラタキクイムシの室内大量飼育法の確立について、その生育と性的成熟における人工飼料用マトリックスの重要性を明らかにし、木粉に換えてセルロースを利用することが可能であることを初めて示した。

２．アフリカヒラタキクイムシの行動の詳細な観察により、雌雄成虫が集合行動を示すことを発見し、雄成虫のヘキサン抽出物が集合フェロモン活性を有することを初めて明らかにした。このヘキサン抽出物の詳細な GC/MS 分析により、主成分 3-pentyl dodecanoate、および 2 つの副成分 2-propyl dodecanoate、3-pentyl tetradecanoate という３エステル化合物が集合フェロモンとして機能することを初めて示した。特に 3-pentyl dodecanoate は天然物として初めての報告である。

３．これらの３成分が天然組成比で混合された場合に最も高い活性を示し、風洞を用いた準実地試験においても十分な効果を有することを明らかにし、アフリカヒラタキクイムシのフェロモンを用いたモニタリングの可能性を見いだした。

以上のように、本論文は外来木材害虫アフリカヒラタキクイムシの生態解明とその防除方法の開発に向け、室内での大量飼育法を確立するとともに、集合フェロモンの発見による新たな防除技術の可能性を見いだしたものであり、木質劣化生物学、木材保存学、昆虫生態学および化学生態学に寄与するところが大きい。

よって、本論文は博士（農学）の学位論文として価値あるものと認める。

なお、平成２７年９月１１日、論文並びにそれに関連した分野にわたり試問した結果、博士（農学）の学位を授与される学力が十分あるものと認めた。

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